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28 STATE STREET  
BOSTON, MA 02109

EXAMINER
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RAO, MANJUNATH N

ART UNIT	PAPER NUMBER
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1652

DATE MAILED: 12/17/2002

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Please find below and/or attached an Office communication concerning this application or proceeding.

# Office Action Summary

Applicati n N .

09/885,297

Applicant(s)

INGRAM ET AL.

Examin r

Manjunath N. Rao, Ph.D.

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-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

## Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133).
- Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

## Status

- 1) ☒ Responsive to communication(s) filed on 10 October 2002.
- 2a) ☐ This action is **FINAL**. 2b) ☒ This action is non-final.
- 3) ☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

## Disposition of Claims

- 4) ☒ Claim(s) 1-110 is/are pending in the application.
- 4a) Of the above claim(s) 1-43,60-96 and 105 is/are withdrawn from consideration.
- 5) ☐ Claim(s) \_\_\_\_\_ is/are allowed.
- 6) ☒ Claim(s) 44-52,54-59,97-102,104 and 106-110 is/are rejected.
- 7) ☒ Claim(s) 53 and 103 is/are objected to.
- 8) ☐ Claim(s) \_\_\_\_\_ are subject to restriction and/or election requirement.

## Application Papers

- 9) ☐ The specification is objected to by the Examiner.
- 10) ☒ The drawing(s) filed on 19 June 2001 is/are: a) ☒ accepted or b) ☐ objected to by the Examiner.
- Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).
- 11) ☐ The proposed drawing correction filed on \_\_\_\_\_ is: a) ☐ approved b) ☐ disapproved by the Examiner.
- If approved, corrected drawings are required in reply to this Office action.
- 12) ☐ The oath or declaration is objected to by the Examiner.

## Priority under 35 U.S.C. §§ 119 and 120

- 13) ☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).
- a) ☐ All b) ☐ Some \* c) ☐ None of:
1. ☐ Certified copies of the priority documents have been received.
2. ☐ Certified copies of the priority documents have been received in Application No. \_\_\_\_\_.
3. ☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).
- \* See the attached detailed Office action for a list of the certified copies not received.
- 14) ☒ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. § 119(e) (to a provisional application).
- a) ☐ The translation of the foreign language provisional application has been received.
- 15) ☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. §§ 120 and/or 121.

## Attachment(s)

- 1) ☒ Notice of References Cited (PTO-892)
- 2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
- 3) ☐ Information Disclosure Statement(s) (PTO-1449) Paper No(s) \_\_\_\_\_.
- 4) ☐ Interview Summary (PTO-413) Paper No(s). \_\_\_\_\_.
- 5) ☐ Notice of Informal Patent Application (PTO-152)
- 6) ☐ Other: \_\_\_\_\_.

### **DETAILED ACTION**

Claims 1-110 are pending in this application. Claims 44-59, 97-104, 106-110 are now under consideration for examination. Claims 1-43, 60-96, and 105 remain withdrawn from consideration as being drawn to non-elected subject matter.

### ***Election/Restrictions***

Applicant's election with traverse of Group III, Claims 44-59, 97-104, 106-110 in Paper No. 9 is acknowledged. The traversal is on the ground(s) that co-examination of all of Groups I-VII would not require independent searches and would not be undue burden to the Examiner. This is not found persuasive because while the searches for some groups appear to overlap, they are not coextensive. The search for Groups I, II and IV to VII would each require the search of subclasses unnecessary for the search of elected Group III. In addition to the patent databases the search also involves extensive non-patent literature. Furthermore contrary to applicant's argument all claims are not linked by a single searchable unifying aspect, i.e., "endoglucanases that degrade oligosaccharides in order to produce ethanol" because applicants have included several other inventions which cannot be determined as novel or not novel simply based on the results of a search on endoglucanases and alcohol production. The requirement is still deemed proper and is therefore made FINAL.

Claims 1-43, 60-96, and 105 are withdrawn from further consideration by the examiner, 37 CFR 1.142(b), as being drawn to a non-elected invention, the requirement having been traversed in Paper No. 9.

***Priority***

Applicant's claim for domestic priority under 35 U.S.C. 119(e) is acknowledged.

***Drawings***

Drawings submitted in this application are accepted by the Examiner for examination purposes only.

***Specification***

The disclosure is objected to because of the following informalities: The specification has blank spaces lacking the ATCC accession number. For example see page 9. Appropriate correction is required.

***Claim Rejections - 35 USC § 112***

The following is a quotation of the second paragraph of 35 U.S.C. 112:

The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

Claim 51 is rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. Claim 51 recites the limitation "said additional enzyme" in 1. There is insufficient antecedent basis for this limitation in the claim.

Claims 106-109 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant

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regards as the invention. Claims 106-109 are drawn to recombinant host strains comprising a specific vector. It is not clear to the Examiner whether the deposits of the bacterial strains were actually made by the applicants because it does not provide the ATCC deposit numbers.

Claims 59 and 106-109 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. Claims 59 and 106-110 are directed to recombinant host strains comprising specific vectors. It is not clear to the Examiner as what is the structure of these vectors and whether they contain specific endoglucanases such as CelY and CelZ along with their respective promoters.

The following is a quotation of the first paragraph of 35 U.S.C. 112:

The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

Claims 59, 106-109 are rejected because the invention appears to employ novel bacterial strains comprising novel vectors. Since the strains/vectors are essential to the claimed invention, they must be obtainable by a repeatable method set forth in the specification or otherwise be readily available to the public. The claimed bacterial strain comprising novel plasmids' sequences are not fully disclosed, nor have all the sequences required for their construction been shown to be publicly known and freely available. The enablement requirements of 35 U.S.C. § 112 may be satisfied by a deposit of the strains or plasmids. The specification does not disclose a repeatable process to obtain the strains or the vectors and it is not apparent if the DNA

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sequences are readily available to the public. Accordingly, it is deemed that a deposit of these plasmids should have been made in accordance with 37 CFR 1.801-1.809.

It is noted that applicants have deposited the organisms but there is no indication in the specification as to public availability. If the deposit was made under the terms of the Budapest Treaty, then an affidavit or declaration by applicants, or a statement by an attorney of record over his or her signature and registration number, stating that the specific strain has been deposited under the Budapest Treaty and that the strain will be irrevocably and without restriction or condition released to the public upon the issuance of the patent, would satisfy the deposit requirement made herein.

If the deposit has not been made under the Budapest treaty, then in order to certify that the deposit meets the criteria set forth in 37 CFR 1.801-1.809, applicants may provide assurance or compliance by an affidavit or declaration, or by a statement by an attorney of record over his or her signature and registration number, showing that:

1. during the pendency of this application , access to the invention will be afforded to the Commissioner upon request;
2. all restrictions upon availability to the public will be irrevocably removed upon granting of the patent;
3. the deposit will be maintained in a public repository for a period of 30 years or 5 years after the last request or for the effective life of the patent, whichever is longer; and
4. the deposit will be replaced if it should ever become inviable.

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Claims 44-53, 55-59 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for a recombinant host cell suitable for degrading oligosaccharide comprising the *celY* encoding polynucleotide isolated from *E.chrysanthemi* as a first heterologous polynucleotide segment encoding a first endoglucanase wherein the segment is under the control of a promoter isolated from fragments of *Sau3A1* gene of *Z.mobilis* (specifically those listed in Table 5 of the specification); and also comprising *celZ* encoding polynucleotide from *E.chrysanthemi* as second heterologous polynucleotide segment under the control of a promoter isolated from fragments of *Sau3A1* gene of *Z.mobilis* (specifically those listed in Table 5 of the specification), wherein said first and second endoglucanase are expressed so that said first and said second degrading activities are present in a ratio that the degrading of said oligosaccharide by said first and second endoglucanase are in synergy, i.e., the sum of the two endoglucanase activities being greater than the sum of individual activities, and such recombinant cell further comprising ethanalogenic enzymes, does not reasonably provide enablement for a recombinant host cell comprising the polynucleotides encoding any two endoglucanases from any source as the first and the second endoglucanases under the control of any promoter from any source and such recombinant cells further comprising additional ethanalogenic enzymes. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to use the invention commensurate in scope with these claims.

Factors to be considered in determining whether undue experimentation is required, are summarized in *In re Wands* (858 F.2d 731, 8 USPQ 2nd 1400 (Fed. Cir. 1988)) as follows: (1) the quantity of experimentation necessary, (2) the amount of direction or guidance presented, (3)

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the presence or absence of working examples, (4) the nature of the invention, (5) the state of the prior art, (6) the relative skill of those in the art, (7) the predictability or unpredictability of the art, and (8) the breadth of the claim(s).

Claims 44-53, 55-59 are so broad as to encompass a recombinant host cell comprising the polynucleotides encoding any two endoglucanases from any source as the first and the second endoglucanases under the control of any promoter isolated from any source and such recombinant cells further comprising additional ethanalogenic enzymes. The scope of the claims is not commensurate with the enablement provided by the disclosure with regard to the extremely large number of DNA sequences that are broadly encompassed by the claims.

The applicants propose to use the above host cells comprising the above polynucleotides (specifically comprising polynucleotides of *E.chrysanthemi* encoding endoglucanases) which they demonstrate to exhibit synergy between each other for the production of ethanol. It appears that invention is solely based on the above phenomenon that inventors have observed with respect to the polynucleotides and the promoters isolated from *E.chrysanthemi*.

Since it appears that the nucleotide sequence of *E.chrysanthemi* alone determines this type of synergy between each other, use of recombinant cells transformed with any polynucleotide encoding endoglucanase isolated from any or all sources as proposed by the applicants may not lead to desired synergy in the host cells. This is because applicants have not shown that using any two polynucleotides encoding any two endoglucanase from any source exhibits such synergistic effects. However, in this case the disclosure is limited to the host cells transformed with endoglucanase encoding polynucleotides from *E.chrysanthemi*.



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While recombinant and mutagenesis techniques are known, it is not routine in the art to screen for multiple substitutions or modifications of nucleotides, as encompassed by the instant claims, and the base changes within a nucleic acid's sequence can be made with a reasonable expectation of success in obtaining the desired activity/utility are limited and the result of such modifications is unpredictable. In addition, one skilled in the art would expect any tolerance to modification for a given DNA to diminish with each further and additional modification, e.g. multiple substitutions.

The specification does not support the broad scope of the claims which encompass host cells transformed with any polynucleotides encoding endoglucanases because the specification does not establish: (A) that synergistic reactions occur between any two endoglucanases isolated from any sources and under the control of any two promoters from any source; (B) regions of the any endoglucanase or its promoter DNA sequence which may be modified to obtain the above mentioned activity/utility; (C) the general tolerance of any endoglucanase encoding DNA or the any promoter DNA sequence to modification and extent of such tolerance; (D) a rational and predictable scheme for modifying any endoglucanase polynucleotide with an expectation of obtaining the desired biological function and utility; and (E) the specification provides insufficient guidance as to which of the essentially infinite possible choices is likely to be successful.

Thus, applicants have not provided sufficient guidance to enable one of ordinary skill in the art to make and use the claimed invention in a manner reasonably correlated with the scope of the claims broadly host cells transformed with any endoglucanase encoding DNA fragment isolated from any source. The scope of the claims must bear a reasonable correlation with the

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scope of enablement (In re Fisher, 166 USPQ 19 24 (CCPA 1970)). Without sufficient guidance, determination of DNAs having the desired biological characteristics is unpredictable and the experimentation left to those skilled in the art is unnecessarily, and improperly, extensive and undue. See In re Wands 858 F.2d 731, 8 USPQ2nd 1400 (Fed. Cir, 1988).

Claims 44-53, 55-59 are rejected under 35 U.S.C. 112, first paragraph, as containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention. These claims are directed to a genus of recombinant cells comprising two heterologous DNA encoding endoglucanases (including their promoters) which exhibit synergy.

The specification does not contain any disclosure of the structure of all DNA sequences encoding endoglucanase (including their respective promoters) which exhibit synergy. The genus of DNAs that comprise these above DNA molecules is a large variable genus with the potentiality of having many different structures. Therefore, many structurally unrelated DNAs are encompassed within the scope of these claims, including partial DNA sequences. The specification discloses only a single species (i.e., host cells transformed with celY and CelZ of *E.chrysanthemi*) of the claimed genus which is insufficient to put one of skill in the art in possession of the attributes and features of all species within the claimed genus. Therefore, one skilled in the art cannot reasonably conclude that the applicant had possession of the claimed invention at the time the instant application was filed.

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Applicant is referred to the revised guidelines concerning compliance with the written description requirement of U.S.C. 112, first paragraph, published in the Official Gazette and also available at [www.uspto.gov](http://www.uspto.gov).

Claims 97-102, 104, 110 are rejected under 35 U.S.C. 112, first paragraph, as containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention. These claims are directed to a genus of recombinant cells comprising two heterologous DNA encoding endoglucanases (including their promoters).

The specification does not contain any disclosure of the structure of all DNA sequences encoding endoglucanase (including their respective promoters). The genus of DNAs that comprise these above DNA molecules is a large variable genus with the potentiality of having many different structures. Therefore, many structurally unrelated DNAs are encompassed within the scope of these claims, including partial DNA sequences. The specification discloses only a single species (i.e., host cells transformed with celY and celZ of *E.chrysanthemi*) of the claimed genus which is insufficient to put one of skill in the art in possession of the attributes and features of all species within the claimed genus. Therefore, one skilled in the art cannot reasonably conclude that the applicant had possession of the claimed invention at the time the instant application was filed.

Applicant is referred to the revised guidelines concerning compliance with the written description requirement of U.S.C. 112, first paragraph, published in the Official Gazette and also available at [www.uspto.gov](http://www.uspto.gov).

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***Claim Rejections - 35 USC § 102***

The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

A person shall be entitled to a patent unless –

(b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

Claims 97-102, 104 and 110 are rejected under 35 U.S.C. 102(b) as being anticipated by Liebl et al. (Microbiology, 1996, Vol. 142(9) :2533-2542). This rejection is based upon the public availability of a printed publication. Claims 97-102, 104 and 110 of the instant application are drawn to a recombinant host cell suitable for degrading an oligosaccharide comprising a first heterologous polynucleotide segment encoding a first endoglucanase and a second heterologous polynucleotide segment encoding a second endoglucanase, suitable for reducing viscosity of an oligosaccharide, wherein the polynucleotide segments are under the transcriptional control of a surrogate promoter, wherein the host cell is a bacterial cell belonging to enterobacteriaceae family, *E.coli* and wherein the first and second endoglucanase is called as EGZ and EGY and wherein the first and the second endoglucanase are sufficiently homologous to the amino acid sequences of celY and celZ from *Erwinia* and share the functional activity of degrading a polysaccharide. Liebl et al. disclose an identical host cell, *E.coli* which comprises two endoglucanases CelA and CelB under the control of a surrogate promoter and which are capable of degrading a polysaccharide. Furthermore, even though the references is not explicit regarding amino acid sequence of the encoded endoglucanases with those of *Erwinia*, Examiner takes the position that they are sufficiently homologous to celY or celZ sequences of *Erwinia*

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based on the functional similarity. Therefore, Liebl et al. anticipate claims 97-102, 104 and 110 as written.

Since the Office does not have the facilities for examining and comparing applicants' protein with the protein of the prior art, the burden is on the applicant to show a novel or unobvious difference between the claimed product and the product of the prior art (i.e., that the protein of the prior art does not possess the same material structural and functional characteristics of the claimed protein). See *In re Best*, 562 F.2d 1252, 195 USPQ 430 (CCPA 1977) and *In re Fitzgerald et al.*, 205 USPQ 594.

### ***Claim Rejections - 35 USC § 103***

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

(a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patent ability shall not be negated by the manner in which the invention was made.

Claims 44-53, 55-58 are rejected under 35 U.S.C. 103(a) as being unpatentable over Riedel et al. (FEMS Microbiology Lett., 1997, Vol. 147:239-243) and Zhou et al. (J. Industrial Microbiol. Biotechnol., 1999, Vol. 22:600-607). Claims 44-53, 55-56, 58 are drawn to a recombinant host cell suitable for degrading an oligosaccharide transformed with a vector expressing two endoglucanases under the control two surrogate promoters in synergy, wherein such recombinant cell secretes endoglucanase, wherein the cell is a bacterial cell belonging to enterobacteriaceae and is either *Escherichia* sp. or *Klebsiella* sp. such as *E.coli* or *K.oxytoca* and wherein the such host cell further comprise additional enzymes as listed in claim 51 and

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additional ethanologenic enzymes and wherein the endoglucanases are called as EGZ or EGY and wherein the additional enzyme a secretory gene product of *pul* or *out* and wherein said cell is ethanologenic.

In summary the above invention appears to be the development of a host cell or a recombinant cell that can be used in the process of simultaneous saccharification and fermentation (SSF). A perusal of the literature indicates that the art is rich in SSF methods and various host cells developed for the above method.

Riedel et al. teach the synergistic interaction of two endoglucanases CelZ and CelY in the degradation of cellulose. However, the reference does not disclose the use of host cell expressing such synergistic endoglucanases. The reference demonstrates the synergistic effects of the two purified enzymes when they are added to the medium containing the cellulose. The reference teaches that the enzymes were purified from culture supernatants of *C.stercorarium* and also provide the methods for its purification. The reference demonstrates the use of two enzymes in synergy leads to a greater level of degradation of cellulose.

Zhou et al. teach the engineering of a *Klebsiella* strain for production of ethanol on an industrial scale using lignocellulosic wastes. After providing the state of the art in engineering ethanologenic microorganisms, the reference teaches methods and also provides a *Klebsiella* strain which has the native ability to metabolize a variety of cellulose substrates and comprising a single endoglucanase derived from *Erwinia* linked to a with a surrogate promoter derived from *Z.mobilis* and additional ethanologenic enzymes. The reference also teaches that the above bacterial strain also has the additional “*out*” gene encoding the type II protein secretion system.

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Thus the above reference teaches all aspects of the above claims except for the fact that the host cell does not have two endoglucanase genes which are synergistic with each other.

Using the teachings of the above two references it would have been obvious to one of ordinary skill in the art to develop a host cell as taught in claims 44-53, 55-56, 58 by first purifying the cDNA encoding the CelZ and CelY taught by Riedel et al. using well known molecular biology techniques. (Techniques to clone the cDNA encoding a purified protein are common knowledge in the art and there are several manual and even commercial Kits available to do the same). Using such clones, it would be obvious to one of ordinary skill in the art to link such endoglucanase sequences to promoter sequences taught by Zhou et al. and transform *Klebsiella oxytoca* P2 strain developed as a biocatalyst for SSF as taught by Zhou et al. One of ordinary skill in the art would have been motivated to do so as there is a great demand for such ethanologenic organisms that are capable of producing ethanol, an industrial solvent and fuel, inexpensively from cellulose wastes. One of ordinary skill in the art would have a reasonable expectation of success since Riedel et al. teach the synergistic effects of CelZ and CelY genes and Zhou et al. provide an ethanologenic strain of *Klebsiella* as well all the methods and promoters for the same.

Therefore the above claims would have been *prima facie* obvious to one of ordinary skill in the art.

This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any

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evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(e), (f) or (g) prior art under 35 U.S.C. 103(a).

### ***Conclusion***

Claims 54 and 103 are objected to as being dependent upon a rejected base claim, but would be allowable if rewritten in independent form including all of the limitations of the base claim and any intervening claims.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Manjunath N. Rao, Ph.D. whose telephone number is 703-306-5681. The examiner can normally be reached on 7.30 a.m. to 4.00 p.m..

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Ponnathapura Achutamurthy can be reached on 703-308-3804. The fax phone numbers for the organization where this application or proceeding is assigned are 703-308-4242 for regular communications and 703-308-4242 for After Final communications.



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Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the receptionist whose telephone number is 703-306-0196.



Manjunath N. Rao, Ph.D.

December 12, 2002

MANJUNATH RAO  
PATENT EXAMINER